

Changes in Firmness of Apples during Exposure to Room Temperature after CA storage

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CA 저장한 사과와 상온방치시 경도변화

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요 약

홍옥과 후지를 3%O₂-2%CO₂ 및 4%O₂-3%CO₂의 CA 저장조건에서 각각 8개월간 저장한 후 20℃, 상대습도 70%의 외기에 방치시키면서 사과의 경도에 관련된 몇가지 변화를 저온 저장고에서 동기간 저장한 후 방치시킨 사과와 비교하였다.

저장후 20℃에 방치한 사과의 총펙틴 함량과 경도는 방치기간이 경과함에 따라 수용성 펙틴의 함량과 polygalacturonase의 활성 증가에 반하여 감소하였다. 이러한 변화는 저온 저장한 사과가 CA 저장한 사과보다 심하였고 홍옥이 후지보다 현저하였는데 특히 방치 초기 5일동안에 더욱 심하였다. 한편 저온저장고에 저장한 홍옥 및 후지는 20℃에서 5일정도 상품적 가치를 지니나 CA 저장한 홍옥은 이보다 5일, 후지는 10일 정도 더 상품성이 연장되는 것으로 관능검사 결과 나타났다.

Introduction

Pectic substances are branched hetero-polysaccharides comprising residues of galacturonic acid, arabinose and galactose with small quantities of other sugars.^{1,2)} Important functions proposed for these components included a major role in cell wall expansion during cell enlargement³⁾ and involvement in textural changes in ripening fruits.^{4,5)} Many of the workers observed the occurrence of pectic compounds in apples as well as the transformation

of the compounds, which take place during storage.⁶⁻¹⁰⁾ In recent years, controlled atmosphere (CA) storage of fruits have been used in commercial purpose. Of the beneficial effects of CA storage on fruits, prevention of ripening processes related to firmness is the most important and apples have been received attention as a material for CA storage. But only few studies on changes in firmness related to pectic substances of fruits during display at normal air after removal from CA storage condition have been carried out.¹²⁻¹⁴⁾

The information on quality changes in fruits during display after CA storage is very important to fruit distributors in the market. For estimation of possible marketing period of apples after removal from CA conditions, some factors related to firmness of apples, such as water-soluble and total pectin and polygalacturonase activity were analyzed, and sensory evaluation on firmness was also carried out in this experiment.

Materials and Methods

Materials and treatments

The apples (Jonathan and Fuji) used in this experiment were grown at the same orchard and manually harvested on Oct. 5 for Jonathan and Oct. 25 for Fuji. The harvested apples were put into wooden boxes and precooled by natural cold air for 5 days before storage.

The fruits were put into the gas tight plastic drums with 100ℓ capacity placed at cold room of $2 \pm 0.2^\circ\text{C}$, $90 \pm 2.0\%$ RH for CA storage of which conditions were 3% O₂-2% CO₂ for Jonathan and 4% O₂-3% CO₂ for Fuji.^{5,16)} The two CA storage conditions had been maintained precisely for 8 months by Lidster's procedure.¹⁷⁾ The apples as a control were also stored at the same cold room. After storage for 8 months the apples stored at the cold room and at the CA storage conditions were exposed to air at $20 \pm 0.5^\circ\text{C}$, $70 \pm 2\%$ RH.

Methods

Preparation of alcohol insoluble solid

Alcohol-insoluble solid was prepared by the procedure of Shewelt.¹⁸⁾ Fifty grams of peeled and sliced apples was added in 200ml of boiling ethanol and heated for 15 min for inactivation of enzymes in flesh. After cooling the mixture was blended with Waring blender for 5 min, and then filtered through Büchner funnel. The

slurry washed twice with 80% ethanol and acetone, then air-dried, weighed and ground to a fine powder.

Extraction and determination of pectic substances

For water-soluble pectin an aliquot of the alcohol-insoluble solid (about 0.1g) was dispersed in 100ml of water by stirring for 30 min at room temperature, and then the supernant was separated by centrifuge at $3,000 \times g$.¹⁹⁾ For determination of total pectin 0.1g of the alcohol-insoluble solid was moistened with 95% ethanol and suspended in 50ml of 0.5% Versenol solution. The mixture pH was adjusted to 11.5 with 1N NaOH and then kept at 30°C for 2 hr for de-esterification. After re-adjustment of the pH to 3.0 with 1N HCl, 100mg of fungal pectinase (Sigma Chemical Co.) and a few drops of toluene were put into the mixture and then the mixture was kept at 25°C overnight. The filtrate from the mixture was separated with filter paper by vacuum. Pectin contents in the supernants extracted as above procedures were measured by the carbazol method.²⁰⁾

Assay of polygalacturonase activity

For preparation of enzyme extract 100g of flesh slices excluded peel was cut from 6 apples, homogenized in 250ml of water and centrifuged at $4,000 \times g$. The residue was suspended in 100ml of 1M NaCl and pH was adjusted to 6.0 with 1M NaOH. The suspension was stirred at 0°C for 12 hr, filtered with cheese cloths and then made to 80% saturated with (NH₄)₂SO₄. The precipitate was collected by centrifuging at $10,000 \times g$, resuspended in 10ml of 0.1M acetate buffer (pH 5.0) and dialysed against the same buffer overnight.

polygalacturonase assay mixture consisted of 0.1ml of the enzyme extract and 4ml of 0.5% polygalacturonic acid (Sigma Chemical Co.) in the acetate buffer was incubated at 25°C for

24 hr. The reaction was terminated with 2N HCl. After centrifugation ($10,000\times g$, 10 min) reducing sugar released was measured by Somogyi method.²¹⁾ Controls in which either the substrate or the enzyme extract were omitted were also included.

The activity of the enzyme was expressed as μg of reducing sugar released by the enzyme under the specific reaction conditions.

Measurement of firmness

Firmness of the apple was evaluated by Terribili's puncture test procedure.²²⁾ Puncture test recordings were taken using a plunger with 8mm diameter mounted on Instron Testing Machine (Model 11140, England), puncturing the flesh at crosshead speed of 100mm/min, reading the maximum force recorded in the first 8mm.

Sensory evaluation on firmness

Qualities on firmness of the uniform and washed apples were scored on hedonic scale by selected 16 members of panel; 9 point was excellent, 5 point was acceptable and 1 point was completely unacceptable. Analysis of variance and mean separation by Duncan's Multiple Range Test ($p. 0.05$)²³⁾ were applied to the data to obtain statistical comparison of treatment.

Results and Discussions

Changes in total pectin content of Fuji and Jonathan apples stored at cold room and at CA storage conditions during display at 20°C are presented in Fig. 1. Total pectin contents at the storage were 0.90% by fresh weight in Jonathan and 1.03% in Fuji. After storage for 8 months the contents reduced to 0.67% in Jonathan and 0.83% in Fuji stored at cold room, and the contents of the apples stored at CA storage conditions were higher than those of the apples stored at cold room.

Total pectin contents in two varieties of apple

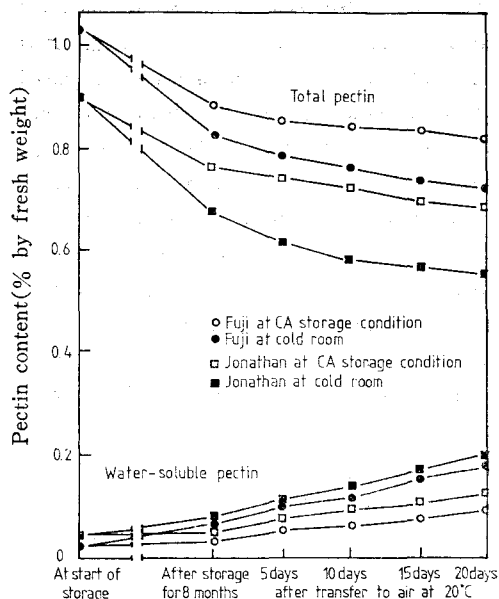


Fig. 1. Changes in total and water-soluble pectin contents of Jonathan and Fuji apples stored at cold room and at CA storage conditions for 8 months and then transferred to air at 20°C

decreased rapidly after transfer to display condition, 20°C, from the storage conditions, which means that vigorous physiological changes occurred in apples owing to the abrupt exposure to high temperature.

Decline of the content in Fuji stored at CA storage condition was slower than in the same apple stored at cold room during display at 20°C. This trend in Jonathan was much clearer than in Fuji.

Fig. 1 also shows that there were considerable variations of water-soluble pectin content in the apples according to varieties and storage conditions. The water-soluble pectin contents were much less than the total pectin content, and the content in Jonathan, 0.02% by fresh weight was higher than in Fuji at start of storage.

After storage and during exposure to 20°C, it decreased with concomitant decline of total pectin content. The increased water-soluble pectin

amount in the two apple varieties was much less than the reduced amount of total pectin. It is supposed that a part of pectic substances degraded from protopectin might be converted into non-pectic substances. In the apples held in CA storage conditions, water-soluble pectin content increased slower than in apples stored at cold room during exposure at 20°C. The content differences between the apples stored at different storage conditions became more pronounced as exposure period progressed.

Fig. 2 shows changes in polygalacturonase activity of the two apple varieties according to the storage conditions during exposure to 20°C after storage. Polygalacturonase activities of the apples were relatively low at the start of storage and increased a little after storage although the total pectin content decreased considerably.

When the apples were exposed to 20°C after storage, the activity increased suddenly. Such

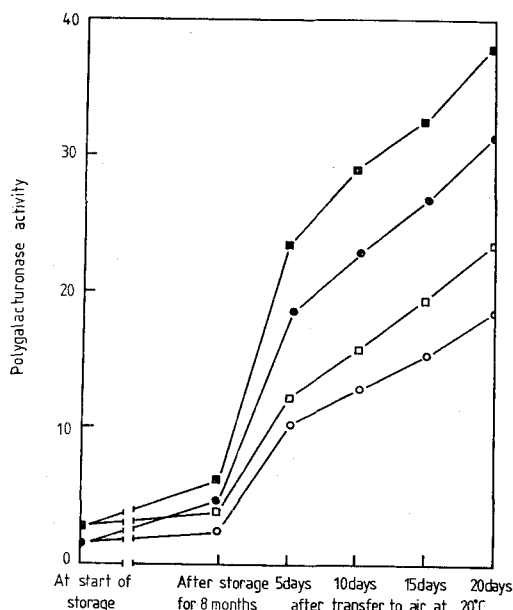


Fig. 2. Changes in polygalacturonase activity of Jonathan and Fuji apples stored at cold room and at CA storage conditions for 8 months and then transferred to air at 20°C

an increase was much more serious in apples stored at cold room than in apples stored at CA storage conditions. This increasing trend was similar to the results on changes in the activity of tomatoes during display at room temperature after removal from the CA storage conditions,¹³⁾ and the reason that the activities in the apples stored at CA storage conditions were lower than in the apples stored at cold room, might be caused by the residual effect of CO₂ gas on the enzyme.

The increased activity during exposure at 20°C after storage is likely due to the formation of inactive insoluble protein to soluble protein caused by intrinsic auto-catalytic processes.²⁴⁾

Fig. 3. presents the changes in firmness of apples during exposure to 20°C after storage. It declined with increase of water-soluble pectin content and Polygalacturonase activity. Tissue softening of fruits related with polygalacturo-

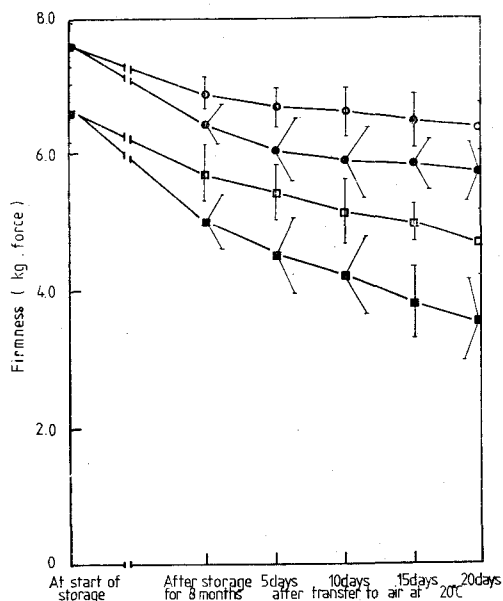


Fig. 3. Changes in firmness of Jonathan and Fuji apples stored at cold room and at CA storage conditions for 8 months and then transferred to air at 20°C
Data points represent mean \pm standard deviation

nase activity was reported by Reymond *et al*²⁵⁾ and the decline of firmness probably results from polygalacturonase activity, for this enzyme hydrolyzes pectin to alcohol-soluble compounds.

After storage firmnesses of the apples stored at cold room reduced by 23.8% in Jonathan and 15.7% in Fuji when compared with the firmnesses of the apples at the start of storage, and it was 8.9% and 6.5% lower than those of Fuji and Jonathan stored at CA storage conditions, respectively.

In exposure of the apples to 20°C, the firmnesses of the apples stored at cold room decreased swiftly for the first 5 days and decline of the firmness in this period was much more serious than any other period. But it was decreased slowly in the apples stored at CA storage conditions.

After exposure for 20 days at 20°C the apples stored at CA storage conditions had higher firmnesses of 30.5% in Jonathan and 12.2% in Fuji than those of the apples stored at cold room.

Table 1 indicates the changes in firmness by organoleptic test of the apples. At the initial storage step the flesh of Fuji was much firmer than Jonathan in mechanical test, but in sensory evaluation, difference in difference in original characteristic in organoleptic aspect acco-

rding to varieties.

Through the changes in total pectin content, water-soluble pectin content and polygalacturonase activity in the apples, the sensory qualities of the two apples decreased after storage. During display at 20°C after storage it was swiftly depreciated in the two varieties, especially in Jonathan stored at cold room.

From the organoleptic test on firmness the apples exposed at 20°C after cold storage had acceptable quality for 5 days after removal from the storage condition. In the firmness by Instron the apples exposed at 20°C for 20 days after CA storage was firmer than the apples exposed for 5 days after cold storage but in sensory evaluation, this apples were acceptable for more than 5 days in Jonathan and more than 10 days in Fuji when compared with the apples stored at cold room.

Abstract

Fuji and Jonathan apples were stored at cold room and at CA storage conditions of 3%O₂-2% CO₂ for Jonathan and 4%O₂-3%CO₂ for Fuji for 8 months, and then transferred to air at 20°C, 70%RH to compare the some factors related to texture of the apples stored at different storage conditions during exposure at 20°C. Total pectin

Table 1. Sensory evaluation on firmnesses of Jonathan and Fuji apples stored at cold room and CA storage conditions and then transferred to air at 20°C

Variety Storage condition Period	Jonathan		Fuji	
	Cold storage	CA storage	Cold storage	CA storage
Start of storage		8.34 ^a		8.56 ^a
Storage for 8 months	7.05 ^c	7.82 ^{ab}	7.41 ^{bc}	8.22 ^a
Storage for 8 months and then 5 days at 20°C	5.12 ^c	6.63 ^b	5.53 ^c	7.47 ^a
and then 10 days at 20°C	3.83 ^d	5.37 ^c	4.75 ^c	6.54 ^a
and then 15 days at 20°C	2.76 ^c	4.65 ^b	4.40 ^b	5.41 ^a
and then 20 days at 20°C	2.05 ^c	3.52 ^b	3.78 ^b	4.65 ^a

* Means within each row followed by the same letter are not significantly different ($p > 0.05$)

content and firmness of the apples reduced considerably with concomitant increases of water-soluble pectin content and polygalacturonase activity during exposure at 20°C after storage.

And the changes were much more serious in the first 5 days than any other period and in the apples stored at cold room, especially in Jonathan than the apples stored at CA storage conditions. From the sensory test the apples stored at cold room maintained acceptable quality for 5 days after removal from storage condition. The acceptability from the test was lengthened for more than 5 days in Jonathan and more than 10 days for Fuji stored at CA storage conditions, when compared with those of the apples stored at cold room.

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